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SEA (SIALYLTRANSFERASE OR GALACTOSYLTRANSFERASE OR FUCOSYLTRANS

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L1 QUE (SIALYLTRANSFERASE OR GALACTOSYLTRANSFERASE OR FUCOSYLTRANS

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FILE 'CAPLUS, EMBASE, BIOSIS, MEDLINE, PASCAL, SCISEARCH, BIOTECHNO,  
JICST-EPLUS, ESBIODASE, TOXCENTER, CANCERLIT, LIFESCI' ENTERED AT  
14:24:29 ON 24 JUN 2003

L2 1572 S L1 AND TRANSMEMBRANE  
L3 227 S L2 AND (DELET? OR DEVOID OR REMOV?)  
L4 76 DUP REM L3 (151 DUPLICATES REMOVED)  
L5 1 S L4 AND ASPERGILLUS

ACCESSION NUMBER: 1992:148892 CAPLUS

DOCUMENT NUMBER: 116:148892

TITLE: The signal for Golgi retention of bovine .beta.1,4-galactosyltransferase is in the transmembrane domain

AUTHOR(S): Teasdale, Rohan D.; D'Agostaro, Giacomo; Gleeson, Paul A.

CORPORATE SOURCE: Med. Sch., Monash Univ., Melbourne, 3181, Australia

SOURCE: Journal of Biological Chemistry (1992), 267(6), 4084-96

CODEN: JBCHA3; ISSN: 0021-9258

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The expression and localization of bovine .beta.1,4-galactosyltransferase (Gal T) was studied in mammalian cells transfected with Gal T cDNA constructs, and the role of N-terminal domains of Gal T in Golgi localization examd. Here it is demonstrated that the transmembrane (signal/anchor) domain of bovine Gal T contains a pos. Golgi retention signal. Bovine Gal T was characterized in transfected cells with antibovine Gal T antibodies, affinity-purified from a rabbit antiserum using a bacterial recombinant fusion protein. These affinity-purified antibodies recognized native bovine Gal T and showed min. cross-reactivity with Gal T from nonbovine sources. Bovine Gal T cDNA was expressed, as active enzyme, transiently in COS-1 cells and stably in murine L cells, and the product was shown to be localized to the Golgi complex by immunofluorescence using the polypeptide-specific antibodies. A low level of surface bovine Gal T was also detected in the transfected L cells by flow cytometry. The removal of 18 of the 24 amino acids from the cytoplasmic domain of bovine Gal T did not alter the Golgi localization of the product transiently expressed in COS-1 cells or stably expressed in L cells. Both the full-length bovine Gal T and the cytoplasmic domain deletion mutant were N-glycosylated in the transfected L cells, indicating both proteins have the correct Nin/Cout membrane orientation. Deletion of both the cytoplasmic and signal/anchor domains of bovine Gal T and incorporation of a cleavable signal sequence resulted in a truncated sol. bovine Gal T that was rapidly secreted (within 1 h) from transfected COS-1 cells. Replacement of the signal/anchor domain of bovine Gal T with the signal/anchor domain of the human transferrin receptor resulted in the transport of the hybrid mol. to the cell surface of transfected COS-1 cells. Furthermore, a hybrid construct contg. the signal/anchor domain of Gal T with ovalbumin was efficiently retained in the Golgi complex, whereas ovalbumin anchored to the membrane by the transferrin receptor signal/anchor was expressed at the cell surface of transfected COS-1 cells. Overall, these studies show that the hydrophobic, signal/anchor domain of Gal T is both necessary and sufficient for Golgi localization.

L4 ANSWER 68 OF 76 BIOSIS COPYRIGHT 2003 BIOLOGICAL ABSTRACTS INC.

ACCESSION NUMBER: 1993:287208 BIOSIS

DOCUMENT NUMBER: PREV199345005333

TITLE: Study of the functional domains of beta-1-4  
**galactosyltransferase** through the expression of the  
**deletion** constructs of cDNA in Escherichia coli and  
mammalian cells: Disulfide bond between CYS 134 and CYS 247  
is required for folding and enzyme activity while the  
**transmembrane** domain is essential for stable  
expression in mammalian cells.

AUTHOR(S): Qasba, Pradman K.; Masibay, Arni S.; Boeggeman, Elizabeth  
E.; Balaji, Peety V.

CORPORATE SOURCE: Div. Cancer Inst., Natl. Inst. Health, Bethesda, MD 20892  
USA

SOURCE: Protein Engineering, (1993) Vol. 6, No. SUPPL., pp. 99.  
Meeting Info.: Winter Symposium on Advances in Gene  
Technology: Protein Engineering and Beyond Miami, Florida,  
USA 1993  
ISSN: 0269-2139.

DOCUMENT TYPE: Conference

LANGUAGE: English

ACCESSION NUMBER: 1993:554766 CAPLUS

DOCUMENT NUMBER: 119:154766

TITLE: Mutational analysis of the Golgi retention signal of bovine .beta.-1,4-**galactosyltransferase**

AUTHOR(S): Masibay, Arni S.; Balaji, Petety V.; Boeggeman, Elizabeth E.; Qasba, Pradman K.

CORPORATE SOURCE: Lab. Math. Biol., Natl. Cancer Inst., Bethesda, MD, 20892, USA

SOURCE: Journal of Biological Chemistry (1993), 268(13), 9908-16

CODEN: JBCHA3; ISSN: 0021-9258

DOCUMENT TYPE: Journal

LANGUAGE: English

AB To examine the role of the N-terminal region of the 402-residue-long acetylglucosamine .beta.-1,4-**galactosyltransferase** (.beta.-1,4-GT), a series of mutants and chimeric cDNA were constructed by polymerase chain reaction and transiently expressed in COS-7 cells, the enzyme activities were measured, and the protein was localized in the cells by subcellular fractionation or indirect immunofluorescence microscopy. It was shown earlier that the **deletion** of the N-terminal cytoplasmic tail and **transmembrane** domain from .beta.-1,4-GT abolishes the stable expression of this protein in mammalian cells. Further **deletion** analyses of the N-terminal region showed that the 1st 21 amino acids of .beta.-1,4-GT are not essential for the stable prodn. of the protein and are consistently localized in the Golgi app. In addn., anal. of hybrid constructs showed that residues 1-25 of .alpha.-1,3-**galactosyltransferase** can functionally replace the .beta.-1,4-GT N-terminal domain (residues 1-43). This fusion protein also showed Golgi localization. On the other hand, the .alpha.-2,6-**sialyltransferase**/.beta.-1,4-GT fusion protein (.alpha.-2,6-ST/.beta.-1,4-GT) needed addnl. C-terminal sequences flanking the **transmembrane** domain of the .alpha.-2,6-ST for stability and Golgi localization. Substitution of Arg-24, Leu-25, Leu-26, and His-33 of the .beta.-1,4-GT **transmembrane** by Ile (pLFM) or substitution of Tyr by Ile at positions 40 and 41 coupled with the insertion of 4 Ile residues at position 43 (pLB) released the mutant proteins from the Golgi and was detected on the cell surface. The results showed that (1) the **transmembrane** domains of .beta.-1,4-GT, .alpha.-1,3-**galactosyltransferase**, and .alpha.-2,6-ST, along with its stem region, all play a role in Golgi targeting and participate in a common mechanism that allows the protein to be processed properly and not be degraded in vivo; (2) increasing the length of the **transmembrane** domain overrides the Golgi retention signal and directs the enzyme to the plasma membrane; and (3) the length of the hydrophobic region of the **transmembrane** domain of .beta.-1,4-GT is an important parameter but is not sufficient by itself for Golgi retention.

ACCESSION NUMBER: 1994:526296 CAPLUS

DOCUMENT NUMBER: 121:126296

TITLE: Expression of soluble active human .beta.1,4-  
**galactosyltransferase** in *Saccharomyces cerevisiae*

AUTHOR(S): Kleene, Ralf; Krezdorn, Christian H.; Watzele, Gabriele; Meyhack, Bernd; Herrmann, Guido F.; Wandrey, Christian; Berger, Eric G.

CORPORATE SOURCE: Physiol. Inst., Univ. Zurich, Zurich, CH-8057, Switz.

SOURCE: Biochemical and Biophysical Research Communications (1994), 201(1), 160-7

CODEN: BBRCA9; ISSN: 0006-291X

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Sequences coding for the cytoplasmic and **transmembrane** domains were **removed** from the cDNA of the human Golgi resident membrane protein .beta.1,4 **galactosyltransferase** (galT). The remaining sequences coding for the stem and catalytic domains of this **glycosyltransferase** were fused to sequences coding for the yeast invertase signal sequence. The hybrid was inserted together with a constitutive yeast promoter and a terminator into an *Escherichia coli*/yeast shuttle vector. *Saccharomyces cerevisiae* strain BT150 transformed with this new expression vector expressed enzymically active sol. enzyme, whereas no activity was detectable in mock-transformed yeasts. The enzyme product was identified by HPLC anal. and shown to correspond to the expected product N-acetyllactosamine.